

Protocol: Heat-shock Transformation

Standard heat-shock transformation of chemically competent bacteria

1. Take competent cells out of -80°C and thaw on ice (approximately 20-30min).
2. Take agar plates (containing the appropriate antibiotic) out of 4°C to warm up to room temperature or place in 37°C incubator.
3. Mix 1 to $5\mu\text{l}$ of DNA (usually 10pg to 100ng) into 20-50 μL of competent cells in a microcentrifuge or falcon tube. GENTLY mix by flicking the bottom of the tube with your finger a few times.
4. Place the competent cell/DNA mixture on ice for 20-30min.
5. Heat shock each transformation tube by placing the bottom 1/2 to 2/3 of the tube into a 42°C water bath for 30-60 seconds (45sec is usually ideal, but this varies depending on the competent cells you are using).
6. Put the tubes back on ice for 2 min.
7. Add 250-500 μl LB or SOC media (without antibiotic) and grow in 37°C shaking incubator for 45min.
8. Plate some or all of the transformation onto a 10cm LB agar plate containing the appropriate antibiotic.
9. Incubate plates at 37°C overnight.